

July 18, 1952

Dr. Philip F. Hartman  
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Dear Dr. Hartman:

Thank you for bringing Blue Tetrazolium (BT) to my attention. I have made some preliminary observations with it which indicate that it will be very satisfactory for my purposes.

I find that the pattern of staining with BT in K-12 is very different from that with Triphenyl tetrazolium (TPT). With TPT, a single polar granule is usually the first object to stain. This may enlarge, and extend axially so as to give the appearance of a deformed club occupying a large part of the interior of the cell. As we have both observed, this granule is not partitioned at cell division. With BT, the staining is more delicate, and many smaller granules appear throughout the cell. This may progress to the extent that only a number of clear spaces (nuclei??) are left. I am not certain, but strongly suspect that these granules are partitioned to the daughter cells about equally. Whether they ~~divide~~ divide individually is another matter. I suspect that the commercial preparation is of questionable purity, and that some difficulties may arise from contamination with inhibitory substances (anisole?). This may account for the apparent sluggishness of its reduction. If a grown culture in broth is used, the reduction appears at least as quickly as with TPT.

It is a very difficult thing to verify, but I suspect that in *E. coli* at least the BT picture is more realistic. Is there independent evidence of such extreme localization of reductive activity in *E. coli* as the TPT picture would suggest?

I tried the same experiment of looking for "non-reducers", with the same unsuccessful result. Earlier, at Stanier's suggestion, I looked into the possibility that the non-aerobic quality of streptomycin-resistant *E. coli*, as reported by Oginsky, was an effect comparable to that of acriflavine on yeast, but could not confirm that streptomycin-resistant mutants were "non-aerobic" even with her strain. Unfortunately, the resistant mutants on which her conclusion was based have been lost.

Yours sincerely,

Joshua Lederberg